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1/26

SEQUENCE LISTING

<110> Eisai Co., Ltd.

<120> CULTURED XENOPUS LAEVIS CELL LINES EXPRESSING MUTANT ADENOMATOUS POLYPOSIS COLI GENE

<130> E1-A0201P

<140>

<141>

<150> JP 2002-241487

<151> 2002-08-22

<160> 9

<170> PatentIn version 3.1

<210> 1

<211> 2829

<212> PRT

<213> Xenopus laevis

<400> 1

Met Ala Ala Ala Ser Tyr Asp Gln Leu Val Lys Gln Val Glu Ala Leu

2 / 2 6

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Thr Met Glu Asn Thr Asn Leu Arg Gln Glu Leu Glu Asp Asn Ser Asn

20               25               30

His Leu Thr Lys Leu Glu Thr Glu Ala Thr Asn Met Lys Glu Val Leu

35               40               45

Lys Gln Leu Gln Gly Ser Ile Glu Asp Glu Ala Met Ala Ser Ser Gly

50               55               60

Pro Ile Asp Leu Leu Glu Arg Phe Lys Asp Leu Asn Leu Asp Ser Ser

65               70               75               80

Asn Ile Pro Ala Gly Lys Ala Arg Pro Lys Met Ser Met Arg Ser Tyr

85               90               95

Gly Ser Arg Glu Gly Ser Leu Ser Gly His Ser Gly Glu Cys Ser Pro

100              105              110

Val Pro Val Gly Ser Phe Gln Arg Arg Gly Leu Leu Asn Gly Ser Arg

115              120              125

Glu Ser Ala Gly Tyr Met Glu Glu Leu Glu Lys Glu Arg Leu Leu Leu

130              135              140

Ile Ala Glu His Glu Lys Glu Glu Lys Glu Lys Arg Trp Tyr Tyr Ala

145 150 155 160

Gln Leu Gln Asn Leu Thr Lys Arg Ile Asp Ser Leu Pro Leu Thr Glu

165 170 175

Asn Phe Ser Met Gln Thr Asp Met Thr Arg Arg Gln Leu Glu Tyr Glu

180 185 190

Ala Arg Gln Ile Arg Ala Ala Met Glu Glu Gln Leu Gly Thr Cys Gln

195 200 205

Asp Met Glu Lys Arg Val Gln Thr Arg Val Gly Lys Ile His Gln Ile

210 215 220

Glu Glu Glu Ile Leu Arg Ile Arg Gln Leu Leu Gln Ser Gln Val Ala

225 230 235 240

Glu Ala Ala Glu Arg Thr Pro Gln Ser Lys His Asp Ala Gly Ser Arg

245 250 255

Asp Ala Glu Lys Leu Pro Asp Gly Gln Gly Thr Ser Glu Ile Thr Ala

260 265 270

Ser Gly Asn Val Gly Ser Gly Gln Gly Ser Ser Ser Arg Ala Asp His

275 280 285

Asp Thr Thr Ser Val Met Ser Ser Asn Ser Thr Tyr Ser Val Pro Arg

290 295 300

Arg Leu Thr Ser His Leu Gly Thr Lys Val Glu Met Val Tyr Ser Leu

305 310 315 320

Leu Ser Met Leu Gly Thr His Asp Lys Asp Asp Met Ser Arg Thr Leu

325 330 335

Leu Ala Met Ser Ser Gln Asp Ser Cys Ile Ala Met Arg Gln Ser

340 345 350

Gly Cys Leu Pro Leu Leu Ile Gln Leu Leu His Gly Asn Asp Lys Asp

355 360 365

Ser Val Leu Leu Gly Asn Ser Arg Gly Ser Lys Glu Ala Arg Ala Ser

370 375 380

Gly Ser Ala Ala Leu Asp Asn Ile Ile His Ser Gln Pro Asp Asp Lys

385 390 395 400

Arg Gly Arg Arg Glu Ile Arg Val Leu His Leu Leu Glu Gln Ile Arg

405 410 415

Ala Tyr Cys Glu Thr Cys Trp Glu Trp Gln Glu Ala His Glu Gln Gly

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420

425

430

Met Asp Gln Asp Lys Asn Pro Met Pro Ala Pro Val Asp His Gln Ile

435

440

445

Cys Pro Ala Val Cys Val Leu Met Lys Leu Ser Phe Asp Glu Glu His

450

455

460

Arg His Ala Met Asn Glu Leu Gly Gly Leu Gln Ala Ile Ala Glu Leu

465

470

475

480

Leu Gln Val Asp Cys Glu Met Tyr Gly Leu Ile Asn Asp His Tyr Ser

485

490

495

Val Thr Leu Arg Arg Tyr Ala Gly Met Ala Leu Thr Asn Leu Thr Phe

500

505

510

Gly Asp Val Ala Asn Lys Ala Thr Leu Cys Ser Met Lys Ser Cys Met

515

520

525

Arg Ala Leu Val Ala Gln Leu Lys Ser Glu Ser Glu Asp Leu Gln Gln

530

535

540

Val Ile Ala Ser Val Leu Arg Asn Leu Ser Trp Arg Ala Asp Val Asn

545

550

555

560

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Ser Lys Lys Thr Leu Arg Glu Val Gly Ser Val Lys Ala Leu Met Glu

565

570

575

Cys Ala Leu Asp Val Lys Lys Glu Ser Thr Leu Lys Ser Val Leu Ser

580

585

590

Ala Leu Trp Asn Leu Ser Ala His Cys Thr Glu Asn Lys Ala Asp Ile

595

600

605

Cys Ser Val Asp Gly Ala Leu Ala Phe Leu Val Ser Thr Leu Thr Tyr

610

615

620

Arg Ser Gln Thr Asn Thr Leu Ala Ile Ile Glu Ser Gly Gly Ile

625

630

635

640

Leu Arg Asn Val Ser Ser Leu Ile Ala Thr Asn Glu Asp His Arg Gln

645

650

655

Ile Leu Arg Glu Asn Asn Cys Leu Gln Thr Leu Leu Gln His Leu Lys

660

665

670

Ser His Ser Leu Thr Ile Val Ser Asn Ala Cys Gly Thr Leu Trp Asn

675

680

685

Leu Ser Ala Arg Asn Ala Lys Asp Gln Glu Gly Leu Trp Asp Met Gly

690

695

700

Ala Val Ser Met Leu Lys Asn Leu Ile His Ser Lys His Lys Met Ile  
705 710 715 720

Ala Met Gly Ser Ala Ala Ala Leu Arg Asn Leu Met Ala Asn Arg Pro  
725 730 735

Ala Lys Tyr Lys Asp Ala Asn Ile Met Ser Pro Gly Ser Ser Val Pro  
740 745 750

Ser Leu His Val Arg Lys Gln Lys Ala Leu Glu Ala Glu Leu Asp Ala  
755 760 765

Gln His Leu Ser Glu Thr Phe Asp Asn Ile Asp Asn Leu Ser Pro Lys  
770 775 780

Thr Thr His Arg Asn Lys Gln Arg His Lys Gln Asn Leu Cys Ser Glu  
785 790 795 800

Tyr Ala Leu Asp Ser Ser Arg His Asp Asp Ser Ile Cys Arg Ser Asp  
805 810 815

Asn Phe Ser Ile Gly Asn Leu Thr Val Leu Ser Pro Tyr Ile Asn Thr  
820 825 830

Thr Val Leu Pro Gly Ser Ser Pro Arg Pro Thr Met Asp Gly Ser

835

840

845

Arg Pro Glu Lys Asp Arg Glu Arg Thr Ala Gly Leu Gly Asn Tyr His

850

855

860

Ser Thr Thr Glu Ser Ser Gly Asn Ser Ser Lys Arg Ile Gly Ile Gln

865

870

875

880

Leu Ser Thr Thr Ala Gln Ile Ser Lys Val Met Asp Glu Val Ser Asn

885

890

895

Ile His Leu Val Gln Glu Asn Arg Ser Ser Gly Ser Ala Ser Glu Met

900

905

910

His Cys Met Ser Asp Glu Arg Asn Ser Gln Arg Lys Pro Ser Ser Asn

915

920

925

His Pro Gln Ser Asn Pro Phe Thr Phe Thr Lys Ala Glu Ser Ser Thr

930

935

940

Arg Gly Cys Pro Val Ala Phe Met Lys Met Glu Tyr Lys Met Ala Ser

945

950

955

960

Asn Asp Ser Leu Asn Ser Val Ser Ser Thr Glu Gly Tyr Gly Lys Arg

965

970

975

9/26

Gly Gln Val Lys Pro Ser Val Glu Ser Tyr Ser Glu Asp Asp Glu Ser

980

985

990

Lys Phe Phe Ser Tyr Gly Gln Tyr Pro Ala Gly Leu Ala His Lys Ile

995

1000

1005

Gln Ser Ala Asn His Met Asp Asp Asn Asp Thr Glu Leu Asp Thr Pro

1010

1015

1020

Ile Asn Tyr Ser Leu Lys Tyr Ser Asp Glu Gln Leu Asn Ser Gly Arg

1025

1030

1035

1040

Gln Ser Pro Thr Gln Asn Glu Arg Trp Ser Arg Pro Lys His Ile Ile

1045

1050

1055

Asp Ser Glu Met Lys Gln Ser Glu Gln Arg Gln Pro Arg Thr Thr Lys

1060

1065

1070

Thr Thr Tyr Ser Ser Tyr Thr Glu Asn Lys Glu Glu Lys His Lys Lys

1075

1080

1085

Phe Pro Pro His Phe Asn Gln Ser Glu Asn Val Pro Ala Tyr Thr Arg

1090

1095

1100

Ser Arg Gly Ala Asn Asn Gln Val Asp Gln Ser Arg Val Ser Ser Asn

1105

1110

1115

1120

1 0 / 2 6

Leu Ser Asn Asn Ser Lys Ala Ser Lys Pro His Cys Gln Val Asp Asp

1125

1130

1135

Tyr Asp Asp Asp Lys Thr Thr Asn Phe Ser Glu Arg Tyr Ser Glu Glu

1140

1145

1150

Glu Gln Gln Glu Asp Glu Thr Glu Arg Gln Asn Lys Tyr Asn Ile Lys

1155

1160

1165

Ala Tyr Ala Ser Glu Glu His His Gly Glu Gln Pro Ile Asp Tyr Ser

1170

1175

1180

Arg Lys Tyr Ser Thr Asp Val Pro Ser Ser Ala Gln Lys Pro Ser Phe

1185

1190

1195

1200

Pro Tyr Ser Asn Asn Ser Ser Lys Gln Lys Pro Lys Lys Glu Gln Val

1205

1210

1215

Ser Ser Asn Ser Asn Thr Pro Thr Pro Ser Pro Asn Ser Asn Arg Gln

1220

1225

1230

Asn Gln Leu His Pro Asn Ser Ala Gln Ser Arg Pro Gly Leu Asn Arg

1235

1240

1245

Pro Lys Gln Ile Pro Asn Lys Pro Pro Ser Ile Asn Gln Glu Thr Ile

1 1 / 2 6

1250

1255

1260

Gln Thr Tyr Cys Val Glu Asp Thr Pro Ile Cys Phe Ser Arg Gly Ser

1265

1270

1275

1280

Ser Leu Ser Ser Leu Ser Ser Ala Glu Asp Glu Ile Glu Gly Arg Glu

1285

1290

1295

Arg Asn Ser Arg Gly Gln Glu Ser Asn Asn Thr Leu Gln Ile Thr Glu

1300

1305

1310

Pro Lys Glu Ile Ser Ala Val Ser Lys Asp Gly Ala Val Asn Glu Thr

1315

1320

1325

Arg Ser Ser Val His His Thr Arg Thr Lys Asn Asn Arg Leu Gln Thr

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1335

1340

Ser Asn Ile Ser Pro Ser Asp Ser Ser Arg His Lys Ser Val Glu Phe

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Ser Ser Gly Ala Lys Ser Pro Ser Lys Ser Gly Ala Gln Thr Pro Lys

1365

1370

1375

Ser Pro Pro Glu His Tyr Val Gln Glu Thr Pro Leu Met Phe Ser Arg

1380

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1390

Cys Thr Ser Gly Ser Ser Leu Asp Ser Phe Glu Ser His Ser Ile Ala

1395 1400 1405

Ser Ser Ile Ala Ser Ser Val Ala Ser Glu His Met Ile Ser Gly Ile

1410 1415 1420

Ile Ser Pro Ser Asp Leu Pro Asp Ser Pro Gly Gln Thr Met Pro Pro

1425 1430 1435 1440

Ser Arg Ser Lys Thr Pro Pro Pro Gln Thr Val Gln Ala Lys Lys

1445 1450 1455

Asp Gly Ser Lys Pro Ile Val Pro Asp Glu Glu Arg Gly Lys Val Ala

1460 1465 1470

Lys Thr Ala Val His Ser Ala Ile Gln Arg Val Gln Val Leu Gln Glu

1475 1480 1485

Ala Asp Thr Leu Leu His Phe Ala Thr Glu Ser Thr Pro Asp Gly Phe

1490 1495 1500

Ser Cys Ala Ser Ser Leu Ser Ala Leu Ser Leu Asp Glu Pro Tyr Ile

1505 1510 1515 1520

Gln Lys Asp Val Gln Leu Lys Ile Met Pro Pro Val Leu Glu Asn Asp

1525 1530 1535

Gln Gly Asn Lys Ala Glu Pro Glu Lys Glu Phe Ile Asp Asn Lys Ala

1540

1545

1550

Lys Lys Glu Asp Lys Arg Ser Glu Gln Glu Lys Asp Met Leu Asp Asp

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1560

1565

Thr Asp Asp Asp Ile Asp Ile Leu Glu Glu Cys Ile Ile Ser Ala Met

1570

1575

1580

Pro Arg Lys Pro Ser Arg Lys Asn Lys Lys Val Pro Gln Pro Thr Pro

1585

1590

1595

1600

Gly Lys Pro Pro Pro Val Ala Arg Lys Pro Ser Gln Leu Pro Val

1605

1610

1615

Tyr Lys Leu Leu Ser Ser Gln Asn Arg Leu Gln Thr Gln Lys His Val

1620

1625

1630

Asn Phe Thr His Ser Asp Asp Met Pro Arg Val Tyr Cys Val Glu Gly

1635

1640

1645

Thr Pro Ile Asn Phe Ser Thr Ala Thr Ser Leu Ser Asp Leu Thr Ile

1650

1655

1660

Glu Ser Pro Pro Ser Glu Pro Thr Asn Asp Gln Pro Asn Thr Asp Ser

1 4 / 2 6

1665

1670

1675

1680

Leu Ser Thr Asp Leu Glu Lys Arg Asp Thr Ile Pro Thr Glu Gly Arg

1685

1690

1695

Ser Thr Asp Asp Thr Asp Ala Ser Lys Pro Leu Asn Pro Thr Thr Val

1700

1705

1710

Leu Asp Glu Asp Lys Ala Glu Glu Gly Asp Ile Leu Ala Glu Cys Ile

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1720

1725

His Ser Ala Met Pro Lys Gly Lys Ser His Lys Pro Tyr Arg Val Lys

1730

1735

1740

Lys Ile Met Asp Gln Ile Asn His Thr Ser Ala Ala Thr Ser Ser Gly

1745

1750

1755

1760

Asn Ser Arg Ser Met Gln Glu Thr Asp Lys Asn Lys Pro Thr Ser Pro

1765

1770

1775

Val Lys Pro Met Pro Gln Ser Ile Gly Phe Lys Glu Arg Leu Lys Lys

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1785

1790

Asn Thr Glu Leu Lys Leu Asn Pro Asn Ser Glu Asn Gln Tyr Cys Asp

1795

1800

1805

1 5/2 6

Pro Arg Lys Pro Ser Ser Lys Lys Pro Ser Lys Val Ala Asn Glu Lys

1810

1815

1820

Ile Pro Asn Asn Glu Glu Arg Thr Lys Gly Phe Ala Phe Asp Ser Pro

1825

1830

1835

1840

His His Tyr Thr Pro Ile Glu Gly Thr Pro Tyr Cys Phe Ser Arg Asn

1845

1850

1855

Asp Ser Leu Ser Ser Leu Asp Phe Glu Asp Asp Asp Ile Asp Leu Ser

1860

1865

1870

Lys Glu Lys Ala Glu Leu Arg Lys Glu Lys Gly Thr Lys Asp Thr Asp

1875

1880

1885

Gln Lys Val Lys Tyr Lys His Glu Asn Arg Ala Ile Asn Pro Met Gly

1890

1895

1900

Lys Gln Asp Gln Thr Gly Pro Lys Ser Leu Gly Gly Arg Asp Gln Pro

1905

1910

1915

1920

Lys Ala Leu Val Gln Lys Pro Thr Ser Phe Ser Ser Ala Ala Lys Gly

1925

1930

1935

Thr Gln Asp Arg Gly Gly Ala Thr Asp Glu Lys Met Glu Asn Phe Ala

1940

1945

1950

1 6 / 2 6

Ile Glu Asn Thr Pro Val Cys Phe Ser Arg Asn Ser Ser Leu Ser Ser

1955

1960

1965

Leu Ser Asp Ile Asp Gln Glu Asn Asn Asn Lys Glu Thr Glu Pro Leu

1970

1975

1980

Lys Gln Thr Gly Thr Ser Glu Thr Gln Leu Gly Leu Arg Arg Pro Gln

1985

1990

1995

2000

Thr Ser Gly Tyr Ala Pro Lys Ser Phe His Val Glu Asp Thr Pro Val

2005

2010

2015

Cys Phe Ser Arg Asn Ser Ser Leu Ser Ser Leu Ser Ile Asp Ser Glu

2020

2025

2030

Asp Asp Leu Leu Gln Glu Cys Ile Ser Ser Ala Met Pro Lys Lys Arg

2035

2040

2045

Lys Pro Ser Lys Ile Lys Asn Glu Val Gly Lys Ser Arg Ser Asn Ser

2050

2055

2060

Val Gly Gly Ile Leu Ala Glu Glu Pro Asp Leu Thr Leu Asp Leu Arg

2065

2070

2075

2080

Asp Ile Gln Ser Pro Asp Ser Glu Asn Ala Phe Ser Pro Asp Ser Glu

1 7 / 2 6

2085

2090

2095

Asn Phe Asp Trp Lys Ala Ile Gln Glu Gly Ala Asn Ser Ile Val Ser

2100

2105

2110

Arg Leu His Gln Ala Ala Ala Gly Ser Leu Ser Arg Gln Gly Ser

2115

2120

2125

Ser Asp Ser Asp Ser Ile Leu Ser Leu Lys Ser Gly Ile Ser Leu Gly

2130

2135

2140

Ser Pro Phe His Leu Thr Leu Asp Lys Glu Glu Lys Thr Ile Thr Ser

2145

2150

2155

2160

Asn Lys Gly Pro Lys Ile Leu Lys Pro Ala Glu Lys Ser Ala Leu Glu

2165

2170

2175

Asn Lys Lys Thr Glu Glu Pro Lys Gly Ile Lys Gly Gly Lys Lys

2180

2185

2190

Val Tyr Lys Ser Leu Ile Thr Gly Lys Ser Arg Ser Ser Ser Asp Phe

2195

2200

2205

Ser Ser His Cys Lys Gln Ser Val Gln Thr Asn Met Pro Ser Ile Ser

2210

2215

2220

1 8 / 2 6

Arg Gly Arg Thr Met Ile His Ile Pro Gly Val Arg Ala Ser Ser Pro  
2225                    2230                    2235                    2240

Ser Thr Ser Pro Val Ser Lys Lys Gly Pro Val Phe Lys Asn Val Pro  
2245                    2250                    2255

Ser Lys Gly Ser Asn Glu Asn Pro Ser Ser Ser Ser Pro Lys Gly  
2260                    2265                    2270

Thr Lys Pro Leu Lys Ser Glu Leu Val Tyr Gly Ser Arg Pro Ser Ser  
2275                    2280                    2285

Thr Pro Gly Gly Ser Ser Lys Gly Asn Ser Arg Ser Gly Ser Arg Asp  
2290                    2295                    2300

Ser Ala Ser Ser Arg Pro Ser Pro Gln Pro Leu Ser Arg Pro Leu Gln  
2305                    2310                    2315                    2320

Ser Pro Gly Arg Asn Ser Ile Ser Pro Gly Lys Asn Gly Ile Ser Pro  
2325                    2330                    2335

Pro Asn Lys Phe Ser Gln Leu Pro Arg Thr Thr Ser Pro Ser Thr Ala  
2340                    2345                    2350

Ser Thr Lys Ser Ser Gly Ser Gly Arg Met Ser Tyr Thr Ser Pro Gly  
2355                    2360                    2365

1 9 / 2 6

Arg Gln Leu Ser Gln Pro Asn Leu Ser Lys Gln Ser Gly Leu Pro Lys

2370                    2375                    2380

Thr His Ser Ser Ile Pro Arg Ser Glu Ser Ala Ser Lys Ser Leu Asn

2385                    2390                    2395                    2400

Gln Asn Val Asn Thr Gly Ser Asn Lys Lys Val Glu Leu Ser Arg Met

2405                    2410                    2415

Ser Ser Thr Lys Ser Ser Gly Ser Glu Ser Asp Arg Ser Glu Arg Pro

2420                    2425                    2430

Ala Leu Val Arg Gln Ser Thr Phe Ile Lys Glu Ala Pro Ser Pro Thr

2435                    2440                    2445

Leu Arg Arg Lys Leu Glu Glu Ser Ala Ser Phe Glu Ser Leu Ser Ser

2450                    2455                    2460

Ser Ser Arg Ala Asp Ser Pro Pro Arg Ser Gln Thr Gln Thr Pro Ala

2465                    2470                    2475                    2480

Leu Ser Pro Ser Leu Pro Asp Met Ala Leu Ser Thr His Ser Ile Gln

2485                    2490                    2495

Ala Gly Gly Trp Arg Lys Met Pro Pro Asn Leu Asn Pro Ala Ala Glu

2 0 / 2 6

2500

2505

2510

His Gly Asp Ser Arg Arg Arg His Asp Ile Ser Arg Ser His Ser Glu

2515

2520

2525

Ser Pro Ser Arg Leu Pro Ile Thr Arg Ser Gly Thr Trp Lys Arg Glu

2530

2535

2540

His Ser Lys His Ser Ser Ser Leu Pro Arg Val Ser Thr Trp Arg Arg

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2550

2555

2560

Thr Gly Ser Ser Ser Ser Ile Leu Ser Ala Ser Ser Glu Ser Ser Glu

2565

2570

2575

Lys Ala Lys Ser Glu Asp Glu Lys Gln Gln Val Cys Ser Phe Pro Gly

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2585

2590

Pro Arg Ser Glu Cys Ser Ser Ser Ala Lys Gly Thr Trp Arg Lys Ile

2595

2600

2605

Lys Glu Ser Glu Ile Leu Glu Thr Pro Ser Asn Gly Ser Ser Ser Thr

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2615

2620

Ile Ala Glu Ser Asn Cys Ser Leu Glu Ser Lys Thr Leu Val Tyr Gln

2625

2630

2635

2640

2 1 / 2 6

Met Ala Pro Ala Val Ser Lys Thr Glu Asp Val Trp Val Arg Ile Glu

2645 2650 2655

Asp Cys Pro Ile Asn Asn Pro Arg Ser Gly Arg Ser Pro Thr Gly Asn

2660 2665 2670

Ser Pro Pro Val Ile Asp Asn Val Leu Asp Gln Gly Gln Lys Glu Glu

2675 2680 2685

Ala Ala Lys Asp Cys His Thr Arg His Asn Ser Gly Asn Gly Asn Val

2690 2695 2700

Pro Leu Leu Glu Asn Arg Gln Lys Ser Phe Ile Lys Val Asp Gly Leu

2705 2710 2715 2720

Asp Thr Lys Gly Thr Asp Pro Lys Ser Leu Ile Asn Asn Gln Gln Glu

2725 2730 2735

Thr Asn Glu Asn Thr Val Ala Glu Arg Thr Ala Phe Ser Ser Ser

2740 2745 2750

Ser Ser Lys His Ser Ser Pro Ser Gly Thr Val Ala Ala Arg Val Thr

2755 2760 2765

Pro Phe Asn Tyr Asn Pro Ser Pro Arg Lys Ser Asn Gly Glu Asn Ser

2770 2775 2780

2 2 / 2 6

Thr Ser Arg Pro Ser Gln Ile Pro Thr Pro Val Thr Asn Ser Thr Lys

2785

2790

2795

2800

Lys Arg Asp Ser Lys Thr Glu Thr Thr Asp Ser Ser Gly Ser Gln Ser

2805

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Pro Lys Arg His Ser Gly Ser Tyr Leu Val Thr Ser Val

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<212> DNA

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<223> an artificially synthesized primer sequence

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23/26

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2 4 / 2 6

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2 5 / 2 6

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19